Salinity Stress

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Influence of Calcium Silicate on Growth, Physiological Parameters and Mineral Nutrition in Two Legume Species Under Salt Stress

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With 3 tables

Received January 25, 2007; accepted July 6, 2007

Abstract

Cowpea and kidney bean plants were grown in a hydroponic system, and the effect of calcium silicate supplied to the nutrient solution under salt stress was investigated. The plants were subjected to four different treatments: (1) nutrient solution alone (C), (2) nutrient solution + 40 mmol 1^{-1} NaCl (NaCl), (3) nutrient solution + 40 mmol l^{-1} NaCl + 0.5 mmol l^{-1} CaSiO₃ (NaCl + Si₁) and (4) nutrient solution + 40 mmol l^{-1} NaCl + 1 mmol l^{-1} $CaSiO_3$ (NaCl + Si₂). The results showed that, in both species, salinity reduced all growth variables but silicate supplementation however partly overcame this growth reduction. Addition of silicate in NaCl-stressed plants maintained membrane permeability. Net photosynthesis, chlorophyll content, stomatal conductance and transpiration were higher in plants under control treatment, and the inclusion of silicate in the nutrient solution resulted in a slight increase in these plant parameters. Intercellular CO₂ was slightly higher in plants under silicate treatment than in plants under control or NaCl treatment. Calcium concentration in shoots and roots in both species was slightly higher in the treatments where silicate was added. Potassium concentration for salt treatment was reduced in shoot and root of both species in the absence of silicate. Sodium and chloride concentration in shoots and roots in both species were slightly higher in the presence of NaCl and were slightly reduced in the plants under silicate treatments. The results suggest that, in hydroponically grown plants, the inclusion of silicate in the nutrient solution is beneficial because it improves growth, physiological parameters and may contribute to a more balanced nutrition by enhancing nutrient uptake under NaCl-stressed conditions. Added calcium silicate may ameliorate the parameters affected by high salinity, may reduce sodium and chloride, and can

slightly increase calcium and potassium concentrations in shoots and roots of salt-stressed cowpea and kidney bean.

Key words: calcium silicate — *Phaseolus vulgaris* — salinity tolerance — *Vigna unguiculata*

Introduction

Salinity is a major stress condition at present (Rueda-Puente et al. 2007) and is one of the most serious environmental problems influencing crop growth (Lopez et al. 2002) and together with drought continues to be one of the world's most serious environmental problems in agriculture. Soil salinity is a major environmental stress that adversely affects plant metabolism and growth. Yield reductions caused by salinity occur on an estimated 30 % of all irrigated land in the United States, with the percentage increasing to 50 % worldwide (Rhoades 1993). Salinity affects plant physiology through changes in the water and ionic status of the cells (Sultana et al. 1999, Hasegawa et al. 2000). Ionic imbalance occurs in the cells due to excessive accumulation of Na⁺ and Cl⁻ which reduces the uptake of other mineral nutrients including Ca^{2+} , Mg^{2+} , Mn and K⁺ (Cramer and Nowak 1992, Khan et al. 1997, Grattan and Grieve 1999, Lutts et al. 1999). Externally supplied Ca^{2+} has been shown to ameliorate the adverse effects of salinity in plant metabolism, presumably by facilitating higher K/Na selectivity (Hasegawa

et al. 2000). Salinity dominated by Na⁺ salts not only reduces Ca2+ availability but also reduces Ca^{2+} transport and mobility to growing regions of the plant, which affects the quality of both vegetative and reproductive organs. Calcium has been shown to ameliorate the adverse effects of salinity on plants (Ehret et al. 1990). Calcium is well known to have regulatory roles in metabolism (Cramer et al. 1986). Sodium ions may compete with calcium ions for membrane-binding sites. Therefore, it has been hypothesized that high calcium levels can protect the cell membrane from the adverse effects of salinity (Busch 1995). On the other hand, silicate has a number of beneficial effects on plant growth under biotic and abiotic stresses (Epstein 1994, 1999, Marschner 1995). Silicon (Si) is not considered an essential element for plant nutrition in the sense of the classical definition of essentiality as postulated by Arnon and Stout (1939). Although Si has been regarded as an essential element in a number of species of the Poaceae and Cyperaceae, it has not been possible to demonstrate, unequivocally, that concentrations of Si above the level of contamination are essential (Epstein 1994). Silicon has frequently been implicated as a 'quasi-essential' element, as an appreciable body of evidence supports the conclusion that Si often enhances plant growth and development (Epstein 1999). Some authors report on lower plant dry matter and yield when growing some plant species in Si-deprived solution, compared to those obtained with solutions containing Si (Mitsui and Takatoh 1963, Chen and Lewin 1969, Miyake and Takahashi 1983, Savant et al. 1997, Epstein 1999).

In other reports, Si was implicated to ameliorate salt stress and determine the possible mechanism governing silicate on some crop species (Adatia and Besford 1986, Matoh et al. 1986, Bradbury and Ahmad 1990, Ahmad et al. 1992, Liang et al. 1996, Yeo et al. 1999, Savvas et al. 2002). Nevertheless, references regarding interactions between salinity and Si in higher plants are limited in the literature.

The objective of this experiment was to test the hypothesis that calcium silicate could enhance salt tolerance in two legumes species, cowpea (*Vigna unguiculata* L. Walp.) and kidney bean (*Phaseolus vulgaris* L.) through improving growth, physiological parameters, and contributing to a more balanced nutrition by enhancing nutrient uptake under NaCl-stressed conditions. Cowpea is an important grain legume crop used as a fodder crop for livestock, as a green vegetable and for dry beans

(West and Francois 1982). It is reported to have a good tolerance to heat and drought (Rachie and Roberts 1974, Vasquez-Tello et al. 1990, Murillo-Amador et al. 2002), and it has a high yield potential under irrigation (Turk et al. 1980). It is grown to obtain seeds and pods for human consumption, as a source of green manure and organic material on unproductive soils, primarily in semi-arid regions. Cowpea has a moderate tolerance to salinity, with a greater tolerance than corn but less than wheat, barley, sugar beet or cotton (Hall and Frate 1996). Kidney bean is a significant source of dietary protein in many developing countries (Durante and Gius 1997). Kidney bean is sensitive to salinity, like many other leguminous crops, and suffers reduced yield even if it is grown at soil salinity less than 2 dS m^{-1} (Maas and Hoffman 1977).

Material and Methods

Plant material and growth conditions

Two independent experiments were conducted and developed at the research facility of the agriculture faculty of Tottori University, Japan. Two legume species, cowpea (cv. UCR CB-27) and kidney bean (cv. Taishokintoki), were transplanted to 41 Wagner pots (Model ICW-1, polycarbonate resin; Fujiwara Scientific Inc., Tsukuba, Japan) under hydroponic and greenhouse conditions and filled with nutrient solution containing the following macro-elements $[(mmol l^{-1}): N, 3.30; P, 0.30; K^+, 1.0; Ca^{2+}, 0.75; Mg^{2+}, 1.0]$ and micro-elements [(mg l⁻¹): Fe, 2.0; Mn, 0.5; B, 0.2; Zn, 0.1; Cu, 0.01; Mo, 0.05] to investigate the effect of calcium silicate supplied to the nutrient solution on plants grown with salt stress. The treatments were applied after the pots were transferred to a naturally illuminated greenhouse and once the primary leaves had developed. Each pot contained five plants, and the nutrient solution was replaced twice per week. The solution was continuously aerated and the pH was adjusted to 5.5, using dilute H₂SO₄ or KOH at the time of renewal.

Treatments

The treatments were (1) nutrient solution alone (C), (2) nutrient solution + 40 mmol l^{-1} NaCl (NaCl), (3) nutrient solution + 40 mmol l^{-1} NaCl + 0.5 mmol l^{-1} CaSiO₃ (NaCl + Si₁) and (4) nutrient solution + 40 mmol l^{-1} NaCl + 1 mmol l^{-1} CaSiO₃ (NaCl + Si₂). The NaCl concentration was gradually increased until it reached the desired concentration (40 mmol l^{-1}).

Physiological variables

Net photosynthesis (Pn, μ mol m² s⁻¹), transpiration rate (E, μ mol m² s⁻¹), stomatal conductance (gs, mmol m² s⁻¹), intercellular CO₂ (Ci, μ mol mol⁻¹) and

chlorophyll (Chl SPAD-502; Minolta Camera Co., Ltd, Osaka, Japan) were measured once a week for 3 weeks on the uppermost, fully-expanded leaves of four plants per treatment. Pn, E, Gs and Ci were measured with a portable monitoring system (ADC, Analytical Development Company, Ltd, type LCA-4, Shimadzu LCA4; Shimadzu Corporation, Kyoto, Japan). At the time of the measurements, mean leaf temperature was 34.5 ± 1.7 and mean photosynthetically active radiation was $753 \pm 62.4 \ \mu mol m^{-2} s^{-1}$.

Electrolyte leakage

Electrolyte leakage (EL) was used to assess membrane permeability. This procedure was based on Lutts et al. (1995). EL was measured, using an electrical conductivity meter (Twin Cond. B-173; Horiba, Ltd, Kyoto, Japan). Leaves from two random plants per replicate were taken from fully expanded leaves. Leaves were cut into 1 cm² segments. Leaf samples were then placed in individual stoppered vials containing 10 ml distilled water after three washes with distilled water to remove surface contamination. These samples were incubated at room temperature (ca. 25 °C) on a shaker (100 r.p.m.) for 24 h. Electrical conductivity of bathing solution (EC_1) was read after incubation. The same samples were then placed in an autoclave at 120 °C for 20 min and the second reading (EC₂) was determined after cooling the solution to room temperature. The EL was calculated as EC_1/EC_2 and expressed as percentage.

Chemical analysis and dry weight determination

Five randomly chosen plants of cowpea and three plants for kidney bean per replicate were divided into shoots and roots. Fresh and dry weight of these tissues was measured. The roots and shoots were washed with three or four rinses in distilled water to remove any dust and other residues and then were dried at 70 °C until constant weight in a circulating oven (ALP Co., Ltd, Tokyo, Japan). The dried tissues were finely ground and stored in paper bags. Chemical analyses were carried out on a dry weight basis. Na⁺, Ca²⁺, Mg²⁺ and K⁺ was determined by atomic absorption spectrophotometer (Shimadzu AA-660; Shimadzu) after digestion with H₂SO₄, HNO₃ and HClO₄. Chloride was extracted in boiling water and the concentration was determined by ion chromatography (Shimadzu HIC-6A; Shimadzu).

Experimental design and statistical analysis

Each treatment at each experiment was arranged in a completely randomized design with four replicates; each replicate included five plants (20 plants per treatment). Data were analysed through ANOVA tests, using ANOVA and GLM procedures in SAS (SAS Institute 1988). Least significant differences among means of species were compared by Tukey's multiple range test at P = 0.05. The EL data were arcsine transformed according to Sokal and Rohlf (1998).

Treatment	Root fresh weight	Root dry weight	Stem dry weight	Shoot dry weight	Whole plant dry weight	Electrolyte leakage
Cowpea ¹						
C	25.60 a	1.82 a	5.13 a	8.91 a	10.74 a	17.91 ± 10.35
NaCl	17.16 b	1.40 b	3.29 b	6.70 b	8.18 b	$27.74~\pm~9.78$
$NaCl + Si_1$	21.90 ab	1.48 ab	3.70 b	7.01 b	8.55 b	23.22 ± 10.86
NaCl+Si ₂	22.13 ab	1.53 ab	3.93 b	7.54 ab	8.94 ab	$20.48~\pm~7.17$
	Leaf dry weight	Root dry weight	Stem dry weight	Shoot dry weight	Whole plant dry weight	Electrolyte leakage
Kidney bean ²						
C	2.01 a	1.00 ns	2.11 a	4.13 a	7.27 ns	15.20 ± 3.61
NaCl	0.90 b	0.49	0.73 b	1.63 b	3.36	35.41 ± 16.13
$NaCl + Si_1$	1.27 ab	0.59	1.20 b	2.48 ab	4.36	28.29 ± 19.17
$NaCl + Si_2$	1.32 ab	0.61	1.21 ab	2.52 ab	4.58	$22.37~\pm~9.17$

Table 1: Effect of calcium silicate on plant growth (g/plant) and electrolyte leakage (%) in cowpea and kidney bean grown in NaCl culture solutions (40 mmol l^{-1})

Mean values followed by the same letter with each column are not significantly different (Tukey P = 0.05). C, control; NaCl, nutrient solution + 40 mmol l⁻¹ NaCl; NaCl+Si₁, nutrient solution + 40 mmol l⁻¹ NaCl + 0.5 mmol l⁻¹ CaSiO₃; NaCl+Si₂, nutrient solution + 40 mmol l⁻¹ NaCl + 1 mmol l⁻¹ CaSiO₃. Each value of electrolyte leakage is the mean \pm S.D. of two leaves per replication. ns, not significant. ¹Mean values of four replicates of five plants per replication.

²Mean values of four replicates of three plants per replication.

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Results

Plant growth

Significant differences between treatments were found in the root fresh and dry weight of cowpea, stem and shoot dry weight of both species, leaf dry weight of kidney bean and whole plant dry weight of cowpea (Table 1). In both cowpea and kidney bean, it is clear that the majority of variables showed higher values in control treatment (nonsaline culture solution). In general, in both species, all growth parameters showed lower values in the NaCl treatment, and afterwards, these growth parameters slightly increased as silicate concentration increased (Table 1).

Electrolyte leakage

Membrane permeability was determined by measuring EL. No significant differences between treatments for either species were present in this variable. However, NaCl treatment impaired membrane permeability by increasing EL. Addition of silicate (NaCl + Si₁ and NaCl + Si₂) maintained membrane permeability, and in general, values of EL were similar between species among treatments, although in kidney bean, EL showed slightly higher values with respect to cowpea under NaCl treatment (Table 1).

Nutrient concentration

As shown in Table 2, calcium concentration showed significant differences in shoots of kidney bean only and it was evident that the concentration was slightly higher in the treatments where silicate was added in roots and shoots in both species. Potassium concentration showed significant differences between treatments in both species, except in shoots of kidney bean. Potassium concentration for salt treatment was reduced in shoots and roots of both species in the absence of silicate compared with the control. Shoot and root potassium concentrations in cowpea for the NaCl + Si₂ treatment were slightly higher than those for the NaCl treatment while in kidney bean no marked differences were observed in root potassium concentration between treatments with salt treatments with silicate compared with salt treatments without silicate. Sodium concentration in both shoots and roots was higher for both species in the presence of NaCl treatment than in the control treatment, and this concentration was slightly reduced in the treatments where silicate was added, therefore, silicate reduced sodium uptake from saline solution into the shoots and roots of both species. In addition, chloride concentration increased in roots and shoots in both species in the presence of NaCl treatment than in the control treatment, but added

Table 2: Effect of calcium silicate on Ca^{2+} , Mg^{2+} , K^+ , Na^+ , and Cl^- concentrations in roots and shoots (mg g⁻¹ dry weight) and ratio of shoot Na to root Na in cowpea and kidney bean grown in NaCl culture solution (40 mmol l^{-1})

Treatment	Concentration in roots				Concentration in shoots				Shoot
	Ca ²⁺	\mathbf{K}^+	Na ⁺	Cl ⁻	Ca ²⁺	\mathbf{K}^+	Na ⁺	Cl ⁻	Na ⁺ /root Na ⁺ ratio
Cowpea ¹									
Ċ	1.21 ns	39.08 a	5.49 b	0.75 b	30.64 ns	69.59 a	18.33 b	2.77 c	3.43 a
NaCl	1.13	22.21 c	37.50 a	28.38 a	33.08	50.26 b	61.40 a	97.43 a	1.89 b
$NaCl + Si_1$	1.43	24.08 c	33.03 a	23.65 a	35.12	56.13 b	56.83 a	76.91 b	1.74 b
$NaCl + Si_2$	1.59	31.12 b	32.71 a	23.18 a	35.44	60.31ab	48.48 a	76.17 b	1.31 b
Kidney bean ²									
C	1.86	29.91 a	3.79 b	2.98 b	31.89 b	93.62 ns	9.18 b	0.91 b	2.49 a
NaCl	1.94	13.29 b	33.85 a	15.49 a	36.91 ab	92.87	41.09 a	120.09 a	1.38 b
$NaCl + Si_1$	2.32	14.38 b	31.44 a	15.04 a	41.64 ab	99.79	31.76 a	112.92 a	1.09 b
$NaCl + Si_2$	2.51	16.77 b	29.32 a	13.21 a	41.69 a	103.81	30.75 a	106.77 a	0.93 b

Mean values followed by the same letter with each column are not significantly different (Tukey P = 0.05). C, control; NaCl, nutrient solution + 40 mmol l^{-1} NaCl; NaCl+Si₁, nutrient solution + 40 mmol l^{-1} NaCl + 0.5 mmol l^{-1} CaSiO₃; NaCl+Si₂, nutrient solution + 40 mmol l^{-1} NaCl + 1 mmol l^{-1} CaSiO₃. ns, not significant.

¹Mean values of four replicates of five plants per replication.

²Mean values of four replicates of three plants per replication.

Table 3: Effect of calcium silicate on net assimilation rate (Pn; μ mol m⁻² s⁻¹), transpiration (E; μ mol m⁻² s⁻¹), stomatal conductance (gs; mmol m⁻² s⁻¹), intercellular CO₂ (Ci; μ mol mol⁻¹), and total chlorophyll (Chl; SPAD-502 readings) of cowpea and kidney bean grown in NaCl culture solution (40 mmol l⁻¹)

Pn	E	gs	Ci	Chl
$8.46~\pm~1.37$	$3.55~\pm~0.80$	$0.37~\pm~0.05$	256.95 ± 26.34	41.34 ± 5.75
$5.96~\pm~1.59$	$3.01~\pm~0.65$	$0.31~\pm~0.12$	288.12 ± 44.90	37.40 ± 5.35
$6.20~\pm~2.31$	$3.13~\pm~0.66$	$0.32~\pm~0.06$	289.47 ± 21.98	$38.49~\pm~7.92$
$6.40~\pm~1.52$	$3.61~\pm~0.37$	$0.39~\pm~0.05$	298.05 ± 34.96	$39.08~\pm~6.00$
$7.56~\pm~2.38$	$3.66~\pm~0.66$	$0.42~\pm~0.13$	275.22 ± 29.88	38.33 ± 1.16
$4.67~\pm~0.48$	$2.23~\pm~0.56$	$0.22~\pm~0.04$	308.27 ± 39.65	$31.08~\pm~3.78$
$5.08~\pm~2.12$	$2.88~\pm~0.51$	$0.30~\pm~0.10$	316.46 ± 45.18	33.62 ± 4.05
$6.23~\pm~0.85$	$2.90~\pm~1.10$	$0.30~\pm~0.08$	378.23 ± 50.91	$33.93~\pm~2.49$
	$8.46 \pm 1.37 5.96 \pm 1.59 6.20 \pm 2.31 6.40 \pm 1.52 7.56 \pm 2.38 4.67 \pm 0.48 5.08 \pm 2.12$	$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	8.46 ± 1.37 3.55 ± 0.80 0.37 ± 0.05 5.96 ± 1.59 3.01 ± 0.65 0.31 ± 0.12 6.20 ± 2.31 3.13 ± 0.66 0.32 ± 0.06 6.40 ± 1.52 3.61 ± 0.37 0.39 ± 0.05 7.56 ± 2.38 3.66 ± 0.66 0.42 ± 0.13 4.67 ± 0.48 2.23 ± 0.56 0.22 ± 0.04 5.08 ± 2.12 2.88 ± 0.51 0.30 ± 0.10	$\begin{array}{c} 8.46 \pm 1.37 \\ 5.96 \pm 1.59 \\ 6.20 \pm 2.31 \\ 6.40 \pm 1.52 \end{array} \begin{array}{c} 3.55 \pm 0.80 \\ 3.01 \pm 0.65 \\ 6.31 \pm 0.12 \\ 0.32 \pm 0.06 \\ 0.32 \pm 0.06 \\ 289.47 \pm 21.98 \\ 298.05 \pm 34.96 \end{array}$

Mean values followed by the same letter with each column are not significantly different (Tukey P = 0.05). C, control; NaCl, nutrient solution + 40 mmol l^{-1} NaCl; NaCl+Si₁, nutrient solution + 40 mmol l^{-1} NaCl + 0.5 mmol l^{-1} CaSiO₃; NaCl+Si₂, nutrient solution + 40 mmol l^{-1} NaCl + 1 mmol l^{-1} CaSiO₃. Each value is the mean \pm S.D. of four plants per treatment and three dates of evaluation.

¹Mean values of four replicates of five plants per replication.

²Mean values of four replicates of three plants per replication.

calcium silicate slightly decreased chloride concentration in shoots and roots of Si-treated plants compared with Si-untreated plants (Table 2).

Physiological variables

The data for physiological variables are shown in Table 3. The net assimilation rate was higher in plants under the control treatment in both species, and this variable was reduced in the NaCl-stressed plants and slightly increased in both treatments where silicate was added. Transpiration in both species was reduced in NaCl-stressed plants, but added silicate increased slightly the transpiration of plants compared with Si-untreated plants. Transpiration in cowpea was slightly higher in the presence of silicate (NaCl + Si_2 treatment) than in plants under control treatment. Stomatal conductance in cowpea was slightly higher under silicate treatment (NaCl + Si_2) while in kidney bean, higher values occurred in plants under the control treatment, reduced in NaCl-stressed plants, but slightly increased in plants under calcium silicate treatments. Intercellular CO_2 in both species was slightly higher in plants under silicate treatments than under NaClstressed and control plants, and higher values occurred in kidney bean. Chlorophyll content (measured indirectly with a SPAD-502) in both species was slightly higher in plants under the control treatment, decreased in NaCl-stressed plants and slightly increased in plants under silicate treatments (Table 3).

Discussion

The results showed that salt stress in both species caused significant reductions in all growth variables, including fresh and dry weight. These findings are in agreement with other reports suggesting that salt stress reduces the biomass of plants of tomato (Navarro et al. 2000, Kaya et al. 2001), cotton (Leidi and Saiz 1997) and rice (Yeo et al. 1999). On the other hand, supplementary silicate resulted in slightly increased plant growth variables (Table 1), where the values were close to those obtained in plants under the control treatment, and in one case (root fresh weight) was slightly higher. Similar results were found in other crops such as rice (Yeo et al. 1999), barley (Okuda and Takahashi 1961, Liang 1999, Ma et al. 2003) and Gerbera (Savvas et al. 2002). The results obtained from EL measurements showed that silicate may act to alleviate salt stress in cowpea and kidney bean by slightly decreasing the permeability of the membrane to help these structures maintain their form (Table 1). Similar results were found by Liang et al. (1996) and Liang (1998).

Net assimilation in cowpea is known to be highly sensitive to salt accumulation in the leaves (Plaut et al. 1990), and consequently the amelioration of sodium uptake by silicate might be expected to enhance photosynthesis. This was investigated and the results showed that the addition of silicate slightly reduced the root and leaf sodium concentration (Table 2) and increased the net assimilation rate and stomatal conductance in both species (Table 3). On the other hand, our results showed that chlorophyll concentration slightly increased in both species in the presence of silicate. These findings are in agreement with reports suggesting that silicate partially offsets the negative impact of NaCl stress, which increased tolerance of tomato plants to NaCl salinity by raising SOD and CAT activities, chlorophyll content and photochemical efficiency of PSII (Khalid Al-aghabary et al. 2004). Transpiration in cowpea is more sensitive to salt accumulation in the leaves than net assimilation rate (Plaut et al. 1990). In this study, transpiration was affected by salinity in both species, but slightly increased in silicate-supplemented plants than in NaCl-stressed plants. The fact that transpiration in both species had slightly increased in plants under silicate treatments and that sodium had slightly decreased in the same treatments, it is clear that silicate did not reduce sodium transport as a consequence of reduced transpiration. This implies that silicate had a direct effect upon sodium transport across the root. Similar results were found in rice by Yeo et al. (1999) and other studies suggested that silicates can complex sodium and prevent its transport into the plant (Matoh et al. 1986, Ahmad et al. 1992). Other studies in non-saline media showed that silica reduced transpiration (Jones and Handreck 1967, Ma and Takahashi 1993), but under saline conditions, any such effect must have been subordinate to effects mediated by reduced sodium concentration in leaves (Yeo et al. 1999). In this study, lower ratio of shoot : root sodium was found in both species, in the presence of calcium silicate (NaCl + Si_2) treatment) (Table 2), suggesting that sodium exclusion was attributed to the presence of silicate, and therefore this exclusion from the shoots is an important mechanism for salt tolerance. Our results are in agreement with Liang (1999) and Forster et al. (1994) in barley, and Gorham et al. (1985) and Greenway and Munns (1980) in Triticeae.

In this study, no significant differences between treatments were found related to concentrations of calcium (except calcium in shoot in kidney bean), but it was evident that plants with silicate treatments exhibited slightly increased calcium concentrations in roots and shoots in both species (Table 2). In studies of rice (Ma and Takahasi 1993), adding silicate decreased shoot calcium and significantly reduced the calcium content of sorghum exposed to aluminium, but had no significant effect on calcium accumulation in plants (Galvez and Clark 1991). In the present study, the fact that both species has slightly increased calcium concentrations when silicate was added may facilitate plants to tolerate salt damage. Similar results were found by Liang (1999) when the barley genotype with tolerance to salinity increased shoot calcium concentration significantly.

On the other hand, silicate-enhanced salt tolerance is believed to be associated with decreased sodium concentration and increased potassium concentration as much in roots and shoots in both species (Table 2). However, the mechanism by which silicate stimulated potassium uptake by inhibiting sodium uptake remains poorly understood, although potassium uptake and transport is an active process associated with ATP-driven H⁺ pump in the plasma membrane (Marschner 1995). According to Liang (1999), one possible mechanism for the stimulating effect of silicate on potassium uptake under NaCl stress is the activation of H⁺-ATPase in the membrane, which was supported by the increased H⁺-ATPase activity in salt-stressed plants in the presence of silicate. None of the mentioned studies related to salinity and silicate in higher plants mentioned the relationship of chloride to silicate. However, in this study, chloride concentration increased in plants under NaCl stress in both species, being higher in kidney bean, but slightly decreased when silicate was present in both species (Table 2). Therefore, chloride uptake and transport into shoots from roots was inhibited by added calcium silicate under stress conditions.

Although the present experiments were conducted on a hydroponic system, other works have demonstrated that the results do not change when the silicate is used under saline stress on a fieldgrown level or a pot experiment using sand, soils or substrate, suggesting that the possible use of SiO_2 for growing plants may be beneficial in areas of high soil salinities (Bradbury and Ahmad 1990), that germination and growth decreased with increasing NaCl concentration in the absence of silicon but the addition of silicon caused significant recovery from salt stress in wheat (Ahmad et al. 1992), and that silicate improves the combined salt and boron tolerance of spinach and tomato grown in naturally sodic-B toxic soil (Aydin et al. 2006). Romero-Aranda et al. (2006) concluded that Si improves the water storage in tomato plants within plant tissues, which allows a higher growth rate that, in turn, contributes to salt dilution into the plant, mitigating the effects of salt toxicity,

while Liang (1999) suggested that silicate is involved in the metabolic or physiological changes in plants (barley) and the optimization of silicate plant nutrition also increases the plant resistance to salt toxicity (Matichenkov and Bocharnikova 2004).

In conclusion, the results of the present work suggest that, in hydroponically grown cowpea and kidney bean crops, the inclusion of calcium silicate in the nutrient solution is beneficial because it improves growth, physiological parameters, and may contribute to a more balanced nutrition by enhancing nutrient uptake under NaCl-stressed conditions. Added calcium silicate may ameliorate the parameters affected by high salinity, may reduce sodium and chloride, and can slightly increase calcium and potassium concentrations in shoots and roots of salt-stressed cowpea and kidney bean. However, and in accord with Liang (1999), further studies are needed for a better understanding of the physiological or biochemical roles of calcium silicate in higher plants.

Acknowledgements

This research was supported by funds of the Consejo Nacional de Ciencia y Tecnología (Project SAGARPA-2004-C01-14), the Programa de Agricultura de Zonas Áridas of the Centro de Investigaciones Biológicas del Noroeste (Project ZA3.1), the Japan International Cooperation Agency and the University of Tottori, Japan.

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